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First Hydroxamic Seconucleoside Derivatives

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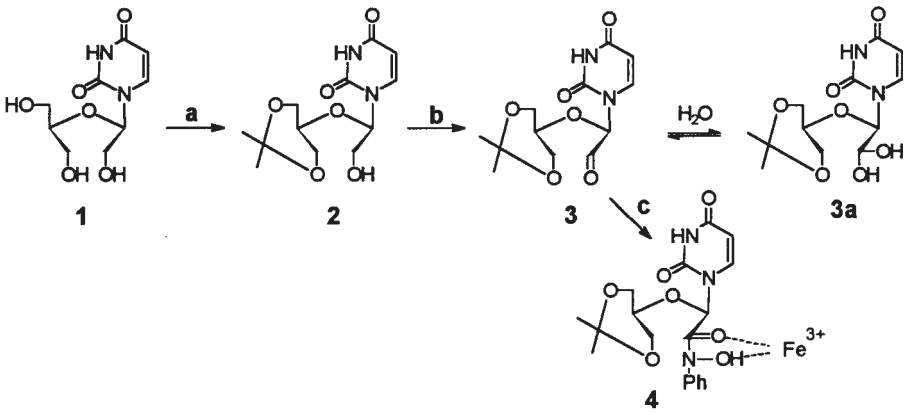
Nucleoside analogues possessing hydroxamate moiety were prepared from secouridine derivatives oxidized at C(2')-position. 3',5'-*O*-Isopropylidene-2',3'-secouridine was oxidized with CrO_3/py complex to the carboxylate level and with DMSO/DCC to the aldehyde level. Both compounds (precursors) gave C(2')-nucleoside hydroxamic acids in the reaction with hydroxylamine or nitrosobenzene in the presence of Fe^{3+} ions, respectively.

A number of hydroxamic acid analogues have been shown to inhibit DNA synthesis by inactivating the enzyme ribonucleotide reductase (RNR).[1–4] This metaloenzyme catalyzes the conversion of (ribo)nucleotides to deoxy(ribo)nucleotides and is therefore a potential target for the development of anticancer agents.[5–8] Hydroxamic acid moiety, R–CONHOH, is found to be the essential pharmacophore in the hydroxyurea, a clinically useful inhibitor of ribonucleotide reductase.[9] A variety of nucleoside analogues are also active as inhibitors of ribonucleotide reductase,[10–12] following the inhibition mechanism similar to that proposed for hydroxamates.[13–15] These results have initiated our interest in the design and synthesis of the nucleoside analogues incorporating hydroxamate moiety. A similiar concept was devised by Farr *et al*.[16] Their compounds inhibited RNR activity, but were 10-fold less potent than hydroxyurea. We, therefore, rationalize that hydroxamate-nucleoside analogues which more closely resemble the RNR substrates should be prepared. Moreover, recent reports[17–19] that hydroxamate compounds increase the potency of nucleosides against HIV-1 give an additional importance to derivatives combining the structural fea-

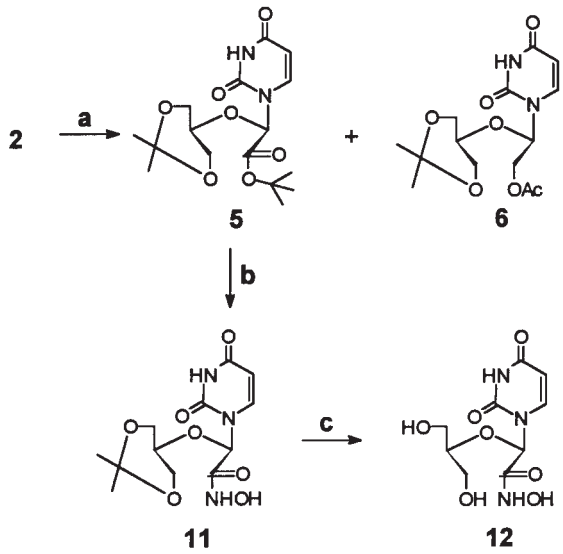
tures of the above compounds. To prepare hydroxamic-nucleoside derivatives, we have used secouridine-2',3'-dialdehyde as the starting material. We have already reported preliminary results on the reaction of nitrosobenzene with 2',3'-secouridine dialdehyde giving hydroxamic-nucleoside product.[20] It is shown that this reaction does not proceed in the absence of ferric ion under the conditions employed. In the presented work, we also prepared monoaldehyde form **3** of 2',3'-secouridine **1** (Scheme 1) in order to prevent the formation of cyclic hemiacetals of dialdehyde monohydrate.[21,22]



Scheme 1. a) acetone, H^+, RT, 2 h; b) DMSO/benzene/pyridine, TFA and DCC, RT, overnight; c) Fe^{3+}, H^+, PhNO in acetone, RT, 6 h.

3',5'-*O*-Isopropylidene-2',3'-seconucleoside **2** was obtained according to the procedure described by Jones *et al*.[23] The oxidation of **2** to the aldehyde level was performed by dimethylsulfoxide (DMSO) and dicyclohexylcarbodiimide (DCC) in the presence of pyridinium trifluoroacetate (PTFA).[24] The aldehyde product **3** was obtained in 60% yield. In the NMR spectra of **3** the signals of aldehyde proton at 10.30 ppm and aldehyde carbon at 194.20 ppm were present, although the compound exists predominately as hydrate **3a**. The test with dinitrophenylhydrazine was positive.

In the reaction of aldehyde **3** with nitrosobenzene, we have observed in UV spectrum the appearance of the highly characteristic peak at 520 nm due to the formation of the iron(III)-mono-*N*-phenylhydroxamate complex **4**. The reaction was promoted both with proton and Fe^{3+} ions. Anyway, the presence of Fe^{3+} in the reaction mixture made it difficult to isolate a free hydroxamic-nucleoside conjugate from complex product **4**. This is due to the known property of hydroxamates to form a strong complex with Fe^{3+}.[25,26] To circumvent this difficulty, we have applied a different approach to the synthesis of hydroxamic-nucleoside derivatives (Scheme 2).

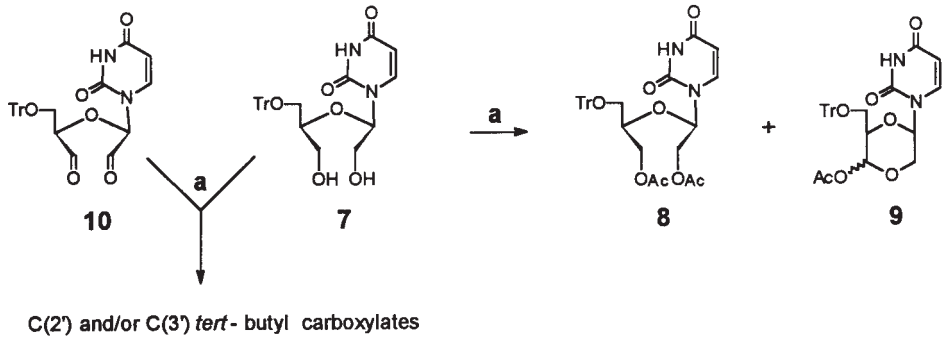


Scheme 2. a) CrO_3/pyridine complex in CH_2Cl_2-DMF, AcOAc, *t*-butanol, RT, 20 h; b) NH_2OH in MeOH, RT, 3 h; c) 40% AcOH, RT, 3 h.

One-step oxidation of primary alcohol **2** to the corresponding carboxylic *tert*-butyl ester **5**[27] was performed using chromium trioxide-pyridine complex[28] in dichloromethane-dimethylformamide (4:1) in the presence of *tert*-butanol and acetic anhydride (60% yield). 10% of the starting compound and 15% of acetylated by-product, 2'-acetyl-3',5'-*O*-isopropylidene-2',3'-secouridine (**6**), were also isolated from the reaction mixture. Omission of dimethylformamide from the reaction mixture led to poorer solubility of the oxidizing complex and consequently to slower oxidation. Therefore, in dichloromethane, the yield of oxidized product **5** was diminished (26%) and the ratio of acetylated product **6** doubled.

5'-*O*-Trityl-2',3'-secouridine **7** (Scheme 3.) could not be oxidized to *tert*--butyl carboxylates at C(2') and/or C(3') position by this method, presumably because of the complex cyclic hemiacetal structures of intermediary dialdehydes.[21,22] The only isolated products were the acetylated starting compound, 2',3'-di-*O*-acetyl-2',3'-secouridine (**8**), and 1-[2(*S*)-trityloxymethyl-3(*R*,*S*)--acetoxy-1,4-dioxan-6(*R*)-yl]uracil (**9**). 1,4-Dioxacyclohexane structure **9** is a diastereomeric mixture resulting from acetylation of the previously formed intramolecular aldehyde hemiacetal. Also, an attempt at oxidation of 5'-*O*--trityl-2',3'-secouridine dialdehyde **10** resulted only in a complex mixture of diastereoisomeric cyclic acetals.

The method used for the preparation of 2'-hydroxamate derivative **11** (Scheme 2.) is similar to that described for the synthesis of the first sugar-



Scheme 3. a) CrO_3/pyridine complex in CH_2Cl_2-DMF, AcOAc, *t*-butanol, RT, 20 h.

hydroxamic acid.[29] Solution of sodium methoxide was added to hydroxylamine hydrochloride to prepare a free hydroxylamine. After removing sodium chloride by filtration hydroxylamine was reacted with *tert*-butyl--3',5'-*O*-isopropylidene-2',3'-secouridine-2'-carboxylate **5** giving 3',5'-*O*-isopropylidene-2',3'-secouridine-2'-hydroxamic acid **11** (65% yield). Deprotection was achieved in 40% acetic acid for 3 hours at room temperature giving hydroxamic-nucleoside derivative **12**[30] (65% yield). As far as we know, it is the first seconucleoside analogue incorporating hydroxamic moiety. We are continuing investigations on hydroxamic-nucleoside analogues with other nucleobases. Metal coordination properties and biological testing of compounds described will be reported in due course.

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27. ^{71}H-NMR ($CDCl_3$) δ/ppm: 9.0 (bs, 1H, NH), 7.51 (d, 1H, J = 8.13 Hz, 6-H), 6.10 (s, 1H, 1'-H), 5.82 (d, 1H, J = 8.14 Hz, 5-H), 4.09–3.70 (m, 5H, 3'-H_2, 4'-H, 5'-H_2), 1.49 (s, 9H, *t*-Bu), 1.42 and 1.41 (2s, 2x3H, *i*Pr). ^{13}C-NMR ($CDCl_3$) δ/ppm: 164.80 (C-2'), 163.42 (C-4), 150.66 (C-2), 140.52 (C-6), 102.82 (C-5), 98.31 (CMe_2), 84.24 (CMe_3), 79.94 (C-1'), 71.60 (C-4'), 61.76 and 61.70 (C-3' and C-5'), 27.51 (CMe_3), 24.16 and 22.20 (CMe_2). *Anal*. Calcd. for $C_{16}H_{24}N_2O_7$ (M_r = 356.38): C 53.93, H 6.79, N 7.86%; found: C 53.98, H 7.03, N 7.71%.
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30. ^{1}H-NMR (DMSO-d_6) δ/ppm: 7.52 (d, 1H, J = 8.0 Hz, 6-H), 6.04 (s, 1H, 1'-H), 5.61 (d, 1H, J = 8.0 Hz, 5-H), 4.0–4.5 (m, NHOH, 2x OH), 3.55–3.33 (m, 5H, 3'-H_2, 4'-H, 5'-H_2).^{13}C-NMR (DMSO-d_6) δ/ppm: 163.73 (C-2'), 161.65 (C-4), 151.29 (C-2), 142.48 (C-6), 101.49 (C-5), 81.92 and 81.40 (C-4' and C-1'), 60.77 and 60.58 (C-3' and C-5'). *Anal*. Calcd. for $C_9H_{13}N_3O_7$ (M_r = 275.22): C 39.28, H 4.76, N 15.27%; found: C 39.33, H 4.75, N 15.30%.

SAŽETAK

Prvi derivati hidroksamskih sekonukleozida

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Nukleozidni derivati s hidroksamskom skupinom pripravljeni su iz sekouridina oksidiranog na položaju C(2'). 3',5'-*O*-Izopropiliden-2',3'-sekouridin oksidiran je s pomoću kompleksa CrO_3/piridin do karboksilata, a pomoću DMSO/DCC do aldehida. Karboksilatni derivat u reakciji s hidroksilaminom, odnosno aldehidni derivat u reakciji s nitrozobenzenom u prisutnosti iona Fe^{3+} daje C(2')-hidroksamski nukleozid.